Antioxidant Activity and Total Phenolic Content of Ethanolic Extracts of Leaf and Seed of *Caesalpinia bonduc*

A Synopsis

Submitted in

partial fulfillment for the degree

of

Doctor of Philosophy

(Biotechnology)

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March, 2012

Introduction

Plants have played a significant role in maintaining human health and improving the quality of human life for thousands of years and have served humans well as valuable components of medicines, seasonings, beverages, cosmetics and dyes. Herbal medicine is based on the premise that plants contain natural substances that can promote health and alleviate illness. In recent times, focus on plant research has increased all over the world and a large body of evidence has collected to show immense potential of medicinal plants used in various traditional systems.

Today, we are witnessing a great deal of public interest in the use of herbal remedies. Furthermore many western drugs had their origin in plant extract. There are many herbs, which are predominantly used to treat cardiovascular problems, liver disorders, central nervous system, digestive and metabolic disorders. Given their potential to produce significant therapeutic effect, they can be useful as drug or supplement in the treatment / management of various diseases. Herbal drugs or medicinal plants, their extracts and their isolated compound(s) have demonstrated spectrum of biological activities. Such have been used and continued to be used as medicine in folklore or food supplement for various disorders. Ethnopharmacological studies on such

Herbs/medicinally important plants continue to interest investigators throughout the world. Caesalpinia bonducella is an Indian herb reported in Ayurveda, the ancient Hindi medicine system of India.

Plants are renewable source of raw materials for the production of variety of chemicals and drugs. Plants produce two broad categories of products; primary and secondary metabolites. Primary metabolites include carbohydrates, lipids and protein while secondary metabolites include alkaloids, steroids, oxygen heterocyclics (flavonoids, anthones, couarnarin), fats, oils, colouring matters etc. These compounds are present in lower volume but possess significant biological activities. The medicinal value of the drug plants is due to the presence of these metabolites, which produce definite physiological action on the human body (Pandey, 1984).

Taxonomy: *Caesalpinia bonduc*

Kingdom: Plantae

Division: Mangoliophyta

Class : Mangoliopsida

Order : Febales

Family: Fabeaceae

Genus : *Caesalpinia*

Species: *Bonduc*

Synonyms:

English: Fever nut, Bonduc nut, Hindi: Kantkarej, Kantikaranja

Habitat: Sandy soils of tropical regions, open, sunny, dry localities; throughout India, in the plains on waste lands and coastal areas.

Distribution: *Caesalpinia bonduc* is pantropical. It is commonly found in coastal areas of tropical Africa.

Plant description: *Caesalpinia bonduc* is pantropical. Liana with stems up to 15 m long, usually armed with robust prickles. Leaves alternate, bipinnately compound, with 6–11 pairs of pinnae; stipules pinnate or 3–5-lobed, up to 20 mm long; petiole and rachis 15–80 cm long; leaflets opposite, 6–9(–12) pairs per pinna, oblong, 2–4 cm × 1–2 cm, base rounded, apex rounded to acute, shortly hairy at least on midrib and margins. Inflorescence a supra-axillary or terminal raceme or panicle 30–60 cm long, densely flowered. Flowers bisexual or functionally unisexual, zygomorphic, 5-merous; sepals free, unequal, c. 5 mm × 2.5 mm, the lowest one hood-shaped; petals free, unequal, 6–7 mm × 2–3 mm, clawed, yellow, the upper one different in shape and size; stamens 10, free, c. 5 mm long, filaments hairy towards base; ovary superior, style short. Fruit an oblong, inflated pod 5–8 cm × 3–4.5 cm, dehiscent, covered with stiff, long hairy prickles, 1–2 seeded. Seeds ovoid, 1.5–2 cm in diameter, smooth, hard, pale grey. Seedling with epigeal germination; cotyledons rounded, thick.

Objectives

- Survey of the fields to select elite mother plants for study.
- Selection of plant parts to standardize the protocol for methanolic and ethanolic extraction.
- To perform the phytochemical analyses of extracts.
- To standardize the protocol for total phenolic content from extract.
- To study the antioxidant activity of methanolic and ethanolic extracts and standardize it’s protocol.

Review of Literature:

Safitri et al., (2003) reported *Antioxidant activity in vitro of two aromatic compounds from Caesalpinia sappan L.* Two antioxidant compounds were isolated from C. sappan L by multiple steps of column chromatography and thin layer chromatography in succession with superoxide scavenging assay as activity monitor. Structures of the two compounds were determined by spectroscopic methods as 1',4'-dihydro-spirobenzofuran-3(2H),3'-3H-2benzopyran-1',6',6',7'-tetrol (compound 1) and 3-4,5-dihydroxy-2 (hydroxymethyl) phenyl-methyl-2,3-dihydro-3,6-benzofurandiol (compound 2). Characterization of antioxidant properties of these two compounds was done by determining the inhibitory effect on xanthine oxidase activity as well as scavenging effect on superoxide anion and hydroxyl radicals.

Datté et al., (2004) reported *Leaf extract of Caesalpinia bonduc Roxb. induces an increase of contractile force in rat skeletal muscle in situ*. The pharmacological properties of Caesalpinia bonduc Roxb. (Caesalpiniaceae) are not well known, but it is used traditionally to treat snake bite (Bellomaria and Kacou, 1995; Schaffner, 1997). In the present study, the mechanism through which Caesalpinia bonduc extract (Cebo) affects gallamine-induced relaxation in rat tibial muscle contractility were studied via measurement of isometric-tension-anesthetized, 10-12-week-old, male rats. Isometric twitch contractions of the indirectly-stimulated anterior tibia muscle of the right hindleg were recorded in situ.

Gupta et al., (2005) reported *Antioxidant Defense System Induced by a Methanol Extract of Caesalpinia bonducella. in Rat Liver*. The antioxidant defense system dramatically controls hepatocellular carcinoma induced by *N*.-nitrosodiethylamine (NDEA). In order to assess the anticarcinogenic activity of a methanol extract of *Caesalpinia bonducella*. (MECB) leaves containing flavonoids and triterpenoids, the antioxidant defense system has been evaluated.

Kumar et al., (2005) reported *Effects of methanol extracts of Caesalpinia bonducella and bauhinia racemosa on hematology and hepatorenal function in mice*. The aim of the present investigation deals with the hematology and hepatorenal function of *Caesalpinia bonducella* Flem. and *Bauhinia racemosa* Lam. The present investigation deals with the sub-chronic toxicity

studies of a methanol extract of *Caesalpinia bonducella* (MECB) leaves and *Bauhinia racemosa* (MEBR) stem bark in Swiss albino mice.

Ali et al., (2008) reported anxiolytic activity of seed extract of *Caesalpinia bonducella (roxb)* in laboratory animals. In the traditional system of Indian medicine *C. bonducella* is widely used for its antipyretic, antiperiodic, anticonvulsant and antiparalytic activities. The present study was aimed to explore the anxiolytic activities of seed extract of *C. bonducella* in experimental animals, mice and rats. In Stair-case model, all the three doses i-e low, medium and high 400, 600 and 800mg/kg of PECB had showed a significant and dose dependent Anxiolytic activity by increasing the number of steps climbed, without any significant effect on rearings by all these three doses.

Ata et al., (2009) reported minor chemical constituents of *Caesalpinia bonduc.* from the ethanolic extract of *Caesalpinia bonduc*, one new diterpene, neocaesalpin P (1) and six known diterpenoids, neocaesalpin H (2), cordylane A (3), caesalpinin B (4), bonducellpin E (5), caesalpinolide A (6), and 17-methylvouacapane-8(14),-9(11)-diene (7) were isolated. Structures of these compounds were determined from NMR spectroscopic studies. Compounds 1-7 exhibited modest antibacterial activities. All of these compounds were weakly active in glutathione S-transferase inhibition assays.

Mandal et al., (2009) gives the assessment of the antioxidant and reactive oxygen species scavenging activity of methanolic extract of *Caesalpinia crista* leaf. Oxidative stress is initiated by reactive oxygen species (ROS), which are responsible for majority of the diseases. However, antioxidants with ROS scavenging ability may have great relevance in the prevention of oxidative stress.

Pawar et al., (2009) reported antioxidant and cytotoxic activities of *Caesalpinia pulcherrima* wood. Antioxidant and cytotoxic activities of the methanolic and aqueous extracts of *Caesalpinia pulcherrima* wood were studied in *in vitro* models. To determine the cytotoxic activity, extracts were tested for toxic effects to brine shrimp larvae. In this assay, the methanolic extract had little effect, but aqueous extract was relatively toxic. The antioxidant and cytotoxic activities may be attributed to the total phenolic content in the wood.

Huda, et al., (2009) reported *Antioxidant activity of plants methanolic extracts containing phenolic* .The presence of natural antioxidant in plants is well known. This paper reports the antioxidative activities of some methanolic plant extracts namely 'ulam raja' (Cosmos caudatus), 'kesum' (Polygonum minus), 'selom' (Oenanthe javanica), 'pegaga' (Centella asiatica) and 'curry leaf' (Murraya koenigii). The analysis carried out was total phenolic content, ferric reducing power, ferric thiocyanate (FTC) and thiobarbituric acid (TBA) tests.

Shukla et al., (2009) has been reported *Antioxidant activity and total phenolic content of ethanolic extract of Caesalpinia bonducella seeds*. This study was to assess the in vitro potential of ethanolic extract of Caesalpinia bonducella seeds as a natural antioxidant. The DPPH activity of the extract (20, 40, 50, 100 and 200 microg/ml) was increased in a dose dependent manner, which was found in the range of 38.93-74.77% as compared to ascorbic acid (64.26-82.58.This study clearly indicate that C. bonducella has a significant potential to use as a natural antioxidant agent.

Almey, et al., (2010) reported *Total phenolic content and primary antioxidant activity of methanolic and ethanolic extracts of aromatic plants' leaves 1.* The aim of this study is to determine the total phenolic content and primary antioxidant activity of methanolic and ethanolic extracts of four aromatic plants' leaves namely knotweed (Polygonum minus), curry (Murraya koenigii), kaffir lime (Citrus hysrix) and fragrant screwpine (Pandanus odurus). Total phenolic content (TPC) assay using Folin-Ciocalteu method was used to assess the presence and level of phenolic compounds in each sample.

Moon et al., (2010) reported *Caesalpinia bonducella F - An Overview.* Many herbal remedies have been employed in various medical systems for the treatment and management of different diseases. This review attempts to encompass the available literature on Caesalpinia bonducella with respect to its pharmacognostic characters, chemical constituents, summary of its various pharmacological activities and traditional uses.

Gill et al., (2011) reported *Phytochemical Investigation of Caesalpinia crista Seed Extract for their Therapeutic Potential***.** Use of plants for health benefits is widespread in India. The present study was to investigate ethanolic seed extract of *Caesalpinia crista* for antioxidant activity by 1,1-diphenyl-2-picryl hydrazyl and hydrogen peroxide methods and anti inflammatory by

Carrageenan induced paw edema and analgesic activity by writhing reflexes and by tail immersion method in mice.

Noorani et al., (2011) reported *Protective Effect of Methanolic Leaf Extract of Caesalpinia Bonduc (L.) on Gentamicin-Induced Hepatotoxicity and Nephrotoxicity in Rats.* The present study was conducted to evaluate the protective effect of methanolic leaf extract of *Caesalpinia bonduc (L.)* on gentamicin-induced hepatotoxicity and nephrotoxicity in rats.

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Methodology

- **Collection of plant material:** Plant materials were collected from our herbal garden in Jayoti Vidyapeeth Women's University, Jaipur, Rajasthan, India. The plant materials were collected in the months of November to February.

- **Preparation of dried plant material:** Leave the plant materials for air dried in shade in Plant tissues culture lab. Dried plant materials are placed in hot air oven for 2 days at 20ºC before formation of powder.

- **Formation of powder:** The dried plant materials were crushed in an electric grinder and pulverized.

- **Preparation of Extract:** Extracts of seed and leaves of *Caesalpinia bonduc* shall be prepared by soxhlet extraction process. The collected residue were preserved in a refrigerator at 2- 8ºC for use in the experiments.

- **Phytochemical investigation:** Phytochemical screening has been done in this phase. Qualitative tests for phytochemical screening of seed and leaf of *C.bonduc* were performed as per the standard methods.

- **Determination of total phenolic content:** Total phenolic content were determined using the Folin-Ciocalteu (FC) reagent method.

- **Assessment of antioxidant activity of methanolic and ethanolic extract:** Antioxidant activity of methanolic and ethanolic extract of seed and leaf of *C. bonduc* shall be assist by various methods.

- **Statistical analysis:** Statistical analysis shall be performed by various softwares according to data.

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